# [COMPOSITIONS AND METHODS FOR PROMOTING NERVE REGENERATION]


## --METHODS OF SCREENING FOR AGENTS THAT PROMOTE NERVE CELL GROWTH--

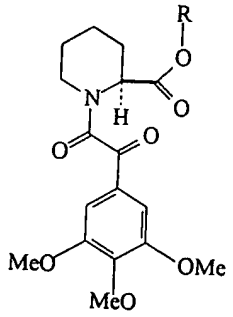
## BACKGROUND OF THE INVENTION

Following traumatic or mechanically induced axonal degeneration in the peripheral nervous system, axonal regeneration ensues, resulting in functional recovery. However, the rate of axonal elongation (3-4 mm/day) is slow. Consequently, recovery is measured in weeks or months, depending upon the distance between the site of injury and the target issue. Therapies that speed regeneration over long distances would be highly beneficial to patients and would significantly reduce health care costs.

The immunosuppressant drug FK506 (SUAN tacrolimus; Prograf®) possesses a variety of neuronal properties, including protection against ischemic brain injury (Sharkey and Butcher, *Nature* **371**:336-339, 1994; Ide et al., *Neurosci. Lett.* **204**:157-160, 1996; Tokime et al., *Neurosci. Lett.* **206**:81-84, 1996) and glutamate toxicity *in vitro* (Dawson et al., *Proc. Natl. Adad. Sci. USA* **90**:9808-9812, 1993), prevention of N-methyl-D-aspartate (NMDA) receptor desensitization (Tong et al., *Science* **267**L1510-1512, 1995), prevention of kindling (Moriwaki et al., *Neurosci. Res.* **25**:191-194, 1996), blockage of long-term potentiation (LTP) and long-term depression (LTD) in the visual cortex (Torii et al., *Neurscience* **185**:1-4, 1995, Funauchi et al., *Neurosci. Res.* **19**269-278, 1994), facilitation of LTP (Ikegami et al., *Mol. Brain Res.* **41**:183-191, 1996) and blockage of LTD in the rat hippocampus (Hodgkiss and Kelly, *Brain Res.* **705**:241-246, 1995), and alternation in neurotransmitter release (Steiner et al., *Mol. Med.* **96**:1076-1151, 1996) and endocytosis (Kuromi et al., *Neurosci. Res.* **27**:101-113, 1997). It is likely that these neuronal properties of FK506 are mediated by calcineurin inhibition. FK506 also speeds functional recovery and axonal regeneration in the rat in a dose-dependent manner following a sciatic nerve crush lesion (Gold et al., *J. Neurosci.* **15**:7505-7516, 1995; Gold et al., *Restor. Neurol. Neurosci.* **6**:287-296, 1994). FK506 was shown to stimulate neuritic outgrowth in a rat pheochromocytoma cell line in a concentration-dependent manner (Lyons et al., *Proc. Natl. Acad. Sci. USA* **91**:3191-3195, 1994).

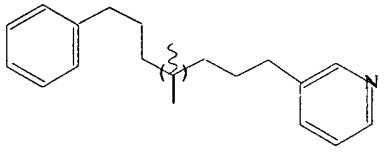
wherein the stereochemistry at position 1 is R or S and the stereochemistry at position 2 is R or S.

(3) Compounds represented by the formula III (see Armistead et al., *Acta. Cryst.* **D51**:522-528, 1995m including a discussion of selection of R and of the synthesis of these compounds, the disclosure of which is incorporated herein by reference):

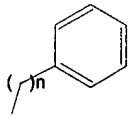



(III)

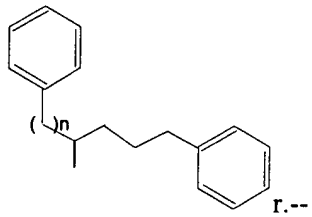
--In one embodiment R is




or

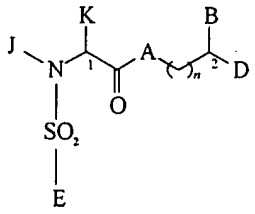


or



r.--

(4) Compounds represented by the formula IV (see WO 92/21313, including a discussion of the synthesis of these compounds, the disclosure of which is incorporated herein by reference):



(IV)

wherein A is $CH_2$, oxygen, NH or N-(C1-C4 alkyl);
wherein B and D are independently Ar, hydrogen, (C1-C6)- straight or branched alkyl or alkenyl that is substituted with a (C5-C7)-cycloalkyl, (C1-C6)-straight or branched alkyl or alkenyl that is substituted with a (C5-C7)-cycloalkenyl, or Ar substituted (C1-C6)-straight or branched alkyl or alkenyl, wherein, in each case, one or two of the $CH_2$ groups of the alkyl or alkenyl chains may contain 1-2 heteroatoms selected from the group consisting of oxygen, sulfur, SO and $SO_2$ in chemically reasonable substitution patterns, or